

one of the most potent endogenous vasodilators in coronary arteries, and appeared to show more potent vasodilating effects on the small-diameter resistant vessels than the large-diameter conduit vessels. The purpose is to study the roles of CGRP in the coronary microcirculation and energy metabolism under ischemic conditions elicited by endothelin-1 (ET-1). Methods: We observed the effects of CGRP on the coronary microcirculation and energy metabolism in isolated beating rat hearts. Microcirculation was observed by an intravital fluorescence videomicroscope system and energy metabolism was evaluated under <sup>31</sup>P-magnetic resonance spectroscopy (<sup>31</sup>P-MRS) after coronary microvessels were pre-contracted with ET-1 (30 pmol), a potent intrinsic vasoconstrictor of small arterioles in myocardium. Results: Cumulative application of CGRP (3–1000 pmol) elicited both dose-dependent reduction in total coronary perfusion resistance (TPR) and simultaneous vasodilation of arterioles of 10–40 µm in diameter (maximal % relaxation was 62%, n = 7). The ED<sub>50</sub> value of CGRP was 30 pmol, about 5000-fold smaller than that of nitroglycerin. Administration of CGRP (100 pmol) decreased TPR (–34%), increased heart rate (+17%), cardiac dP/dt (+76%), work index (+57%), ATP (+21%) and high-energy phosphates (PCr) (+45%) significantly (p < 0.05, n = 14) in myocardium. Conclusion: CGRP relaxed the coronary microvessels, improved the myocardial high-energy metabolism and enhanced the cardiac contractility in the ischemic myocardium, suggesting that the CGRP may play beneficial roles in myocardial ischemia elicited by ET-1.

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### The role of endothelin receptors (ETRA/B) in fibrocyte differentiation

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Introduction: Scleroderma (SSc) is an autoimmune connective tissue disease of unknown aetiology. Pulmonary involvement including the development of pulmonary arterial hypertension (PAH) is characterised by vascular remodelling, collagen deposition and expression of connective tissue growth factor (CTGF). CD14+ monocytes can differentiate into spindle shaped cells termed 'fibrocytes'. Fibrocytes express haematopoietic and mesenchymal markers including collagen, and amplify inflammatory/immune responses via antigen presentation and chemokine secretion. Fibrocyte differentiation is enhanced by fibrogenic cytokines including PDGF. The role fibrocytes play in promoting PAH in SSc is unknown. Methods: CD14+ PBMCs were isolated from SSc and healthy donor blood. Fibrocyte differentiation in the presence of MCSF and/or ET-1 was assessed after 14 days. The effect of endothelin receptor (ETR) antagonists (selective/dual) on fibrocyte differentiation (n = 6) was investigated. SSc and control fibrocyte secretomes were assessed by ELISA (n = 6), and the effects on fibroblast-mediated gel contraction determined. Results: MCSF and ET-1 alone and in combination induced fibrocyte differentiation (P < 0.05). SSc fibrocytes exhibited enhanced differentiation from CD14+ PBMCs than healthy control donors in response to MCSF (P < 0.05), ET-1 (P < 0.05) and in combination (P < 0.01). ETR antagonists BQ123 (ETRA), BQ788 (ETRB) and Bosentan (ETRA/B) inhibited MCSF induced fibrocyte differentiation. CTGF secretion was elevated in SSc compared to control fibrocytes (P < 0.05) cultured with MCSF.

Conditioned media from SSc fibrocytes promoted gel contraction by control pulmonary fibroblasts (P < 0.05). Discussion CD14+ SSc PBMCs readily differentiate into fibrocytes in response to ET-1 and MCSF via ETRA and ETRB. Our data suggests that fibrocytes contribute to the development of PAH in SSc via a paracrine mechanism modulating the functional activities of resident tissue fibroblasts.

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### Improved relaxations to acetylcholine in murine carotid arteries with heterozygous overexpression of preproendothelin-1 in the endothelium

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The endothelium can release both NO and contracting factors (EDCFs). Exogenous endothelin-1 (ET-1) causes ETB receptor mediated release of NO, but also enhances endothelium-dependent contractions. Besides its propensity to exhibit EDCF-mediated contractions, the murine carotid artery is characterized by high basal and stimulated NO generation. The role of the endothelial endothelin system on endothelium-dependent relaxations in this preparation is unknown. Therefore, a model of endothelium-restricted heterozygous overexpression of ppET-1 was used (TET +/– mice). Relaxations were studied and compared in carotid arteries of 34–36 weeks old TET +/– mice and WT littermates. Experiments were performed, in the presence of meclofenamate to exclude endothelium-dependent contractions, in rings suspended in Halpern–Mulvany myographs. Responses to phenylephrine (1 nM to 30 µM) were similar between genotypes, and the final levels of contraction were not significantly different (57 ± 6% KCl in WT vs. 49 ± 5% KCl in TET +/–). Acetylcholine-induced relaxations were potentiated in TET +/– mice compared to littermate controls (PD2 8.37 ± 0.05 vs. 8.61 ± 0.06 in TET +/–, n = 7–10, P < 0.01). By contrast, endothelium-independent relaxations to sodium nitroprusside were not different (n = 6–8). In the presence of meclofenamate, TET +/– had no effect on contractions to the calcium ionophore A23187 (n = 6–7), but maximal responses to the TP receptor agonist U46619 (0.1 nM to 3 µM) were decreased compared to WT control mice (Emax 123.4 ± 3.5% vs. 108.1 ± 2.5% KCl in TET +/–, n = 6–9, P < 0.01). These results suggest that moderate increases in endothelial ET-1 expression in murine carotid arteries enhance endothelium-dependent, NO-mediated relaxations and reduce smooth muscle responsiveness to TP receptor activation.

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### Vascular pharmacology of quercetin in rat

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Quercetin, a kind of flavonoids, exerts the cardiovascular actions. In rat aorta, quercetin (0.1 to 100 µM) relaxed the contraction induced by